BP17

GENOMIC CHARACTERIZATION OF *SUGARCANE MILD MOSAIC VIRUS*

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Sugarcane mild mosaic virus (SCMMV) that was first discovered by Lockhart et al. (1992) was provisionally assigned to the genus *Ampelovirus*, family *Closteroviridae* (Martelli et al 2002). Since the initial characterization of SCMMV using enzyme immunosorbent assay and immunosorbent electron microscopy, no new information about SCMMV has been obtained. Recently, without a priori metagenomics-based approaches (including virion-associated nucleic acid and siRNAs sequencing) were used for viral screening of sugarcane varieties from sugarcane quarantine and germplasm collections. The combination of both metagenomics approaches yielded 918 contigs sharing homologies with large parts of the genome of representative members of the *Ampelovirus* genus and potentially covering 80.6% of typical full-length ampelovirus genomes. In addition, RNASeq or whole transcriptome shotgun sequencing approach based on total RNA extracted from an ampelovirus infected sugarcane variety yielded a 12,408 nt long scaffold of SCMMV. Resequencing PCR, including a gene walking approach and RACE PCRs yielded the full-length genome sequence of SCMMV with a size of 13,144 nt. The most closely related ampelovirus to the novel ampelovirus is *Plum bark necrosis stem pitting-associated virus* with only 43.4% identity, suggesting, as expected, that the agent identified is a novel ampelovirus. High-throughput sequencing data were used to design specific detection primers located within the HSP70 gene for diagnosis. Using these primers, 16 % of sugarcane varieties from the CIRAD sugarcane quarantine program tested positive for SCMMV, providing a first vision of

the geographic distribution (Argentina, Barbados, Ecuador, Guadeloupe, Philippines, Réunion, Senegal, and USA), prevalence, and diversity of this virus. Phylogenetic analysis of the HSP70 gene sequence grouped these isolates into four genetic groups. Immuno-capture PCR using SCMMV antibodies developed by B.E.L. Lockhart detected the three genetic groups for which single strain infection was available, confirming that SCMMV and the newly identified ampelovirus are the same entity.

Lockhart, B. E. L., Autrey, L. J.-C., and Comstock, J. C. 1992. Phytopathology 82:691-695

Martelli G. P., Agranovsky A. A., Bar-Joseph M., Boscia D., Candresse T., et al. 2002.Arch. Virol. 147:2039-44.